UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

OFFICE OF
PREVENTION, PESTICIDES AND
TOXIC SUBSTANCES

Friday, June 26, 2009

MEMORANDUM

Subject: Acute Toxicity Review for EPA Reg. No.: 10324-RIL
Product Name: Maquat 25:12
DP Barcode: D364459

From: Ian Blackwell, Biologist IB
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To: Marshall Swindell, PM 33/ Demson Fuller
Regulatory Management Branch
Antimicrobials Division (7510P)

Applicant: Mason Chemical Company

FORMULATION FROM LABEL:

Active Ingredient(s):		% by wt.
Glutaraldehyde		25.7
Alkyl dimethyl benzyl ammonium chloride		5.0
Didecyl dimethyl ammonium chloride		7.5
Other Ingredient(s):		61.8
	Total:	100.0

1) BACKGROUND: The Mason Chemical Company has submitted a set of three acute toxicity studies to support the data requirements of their product, "Maquat® 25:12". The three studies were conducted by Tox Monitor Laboratories.

The Product Science Branch (PSB) /Antimicrobials Division (AD) contractor, Computer Sciences Corporation (CSC), conducted a primary review of these studies. The Chemistry and Toxicology Team (CTT) conducted a brief secondary review to assure that the studies meet EPA/OPP criteria.

This submission includes a data matrix dated 3/20/2009 data matrix for Maquat 25:12. It states that the Mason Chemical Company intends to use the Cite-All method to support the data requirements for acute inhalation toxicity, primary eye irritation and primary skin irritation. PM Team 33 proposed EPA Registration Number 59894-4 as a product which might have acute toxicity data that could be cited in support of 10324-RIL. Both products contain approximately 50% Glutaraldehyde and approximately 12.5% quaternary ammonium compounds.

CTT searched Agency files for acute toxicity information on 59894-4 and to obtain Confidential Statements of Formula (CSF). A November 29, 1990 data matrix for 59894-4(U) states that the registrant intended to use public literature to support the six acute toxicity study requirements. No other acute toxicity data or reviews were found for 59894-4.

2) RECOMMENDATIONS: PSB findings are:

a) The acute oral toxicity study is Acceptable.
b) The acute dermal toxicity study is Acceptable.
c) The dermal sensitization study is graded Supplementary. This study's issue is a reporting error. The report's table of contents refers to a historical positive control study; however, there is no such data included in that report. Mason says this data is available. However, Mason must amend the dermal sensitization report to properly include that data.
d) CTT is not able to locate any acceptable data to support the acute toxicity data requirements for 10324-RIL. Thus, there is a data gap for the acute inhalation toxicity, primary eye irritation and primary skin irritation studies. Mason must find some way to address the data gaps for 10324-RIL.

DATA REVIEW FOR ACUTE ORAL TOXICITY TESTING (OPPTS 870.1100)
(UP AND DOWN PROCEDURE)

Product Manager: 33
MRID No.: 477075-03

Reviewer: CSC and Ian Blackwell
Completion Date: February 3, 2009
Project ID: 08-124-3

Testing Laboratory: Tox Monitor Laboratories, Inc., Oak Park, IL
Author: Michael Kukulinski, B.S., L.A.T.G.

Quality Assurance (40 CFR §160.12): A Quality Assurance (QA) statement was included. A statement of Good Laboratory Practice (GLP) compliance was included stating that this study was conducted in accordance with the U.S. EPA Good Laboratory Practice Standards, 40 CFR 160.

Test Material: Maquat 25:12, 25% Glutaraldehyde, 12.5% active quat
Lot #: 1621-224 / Clear liquid

Dosage: Limit Test: 5,000 mg/kg (administered neat)
Main Test: 175 mg/kg, 550 mg/kg, and 1,750 mg/kg (administered neat)

Species: 10 Rats; Sprague-Dawley derived, albino
Sex: Females. Females were nulliparous and non-pregnant.
Age: Young adult (8-12 weeks old)
Weight: 177-246 grams
Source: Harlan Sprague Dawley, Indianapolis, IN
Housing: Temperature Range: 66-77°F (19-25°C)
Humidity Range: 30-70%
Photoperiod: 12-hour light/12-hour dark cycle
Acclimation: At least five days

Conclusion:

1. **Acute Oral LD_{50} (mg/kg)**: Female Rats: 550 mg/kg
~95% Confidence Interval: 215.9-1,140.0 mg/kg

2. **Toxicity Category**: III **Classification**: Acceptable

Procedure (Deviations from 870.1100):

- The guidelines state that body weight changes should be calculated and recorded. Individual body weights of test animals were recorded; however, body weight changes were not reported.

The acute toxicity profile for Registration Number 10324-RIL is currently:

Study	MRID Number	Toxicity Category	Status
Acute Oral Toxicity	477075-03	III	Acceptable
Acute Dermal Toxicity	477075-04	III	Acceptable
Acute Inhalation Toxicity		?	Data Gap
Primary Eye Irritation		?	Data Gap
Primary Skin Irritation		?	Data Gap
Dermal Sensitization	477075-05	?	Supplementary

3) LABELING:

a) Due to the three data gaps, no precautionary labeling can be prescribed at this time.

Results:

Limit Test

Dosing Sequence	Animal No.	Dose Level (mg/kg)	Short-Term Outcome	Long-Term Outcome
1	690	5,000	D	D

D – Death

Main Test

Dosing Sequence	Animal No.	Dose Level (mg/kg)	Short-Term Outcome	Long-Term Outcome
1	696	175	S	S
2	698	550	S	S
3	700	1,750	D	D
4	702	550	D	D
5	703	1,750	D	D
6	722	550	D	D
7	779	175	S	S
8	780	550	S	S
9	781	1,750	D	D

S – Survival; D – Death

Observations:

5,000 mg/kg Dose Level (1 animal): This animal died within one day of test substance administration. Prior to death, the animal was hypoactive and exhibited a state of extreme bodily exhaustion.

175 mg/kg Dose Level (2 animals): Both animals survived, gained body weight, and appeared active and healthy during the study.

550 mg/kg Dose Level (4 animals): Two animals died and two animals survived at this dose level. The two animals that survived gained body weight by Day 14 and appeared active and healthy during the study, except for piloerection that was noted in both surviving animals through Day 1. The animal that died on Day 1 exhibited piloerection and hypoactivity prior to death. The animal that died on Day 7 exhibited piloerection, hypoactivity, and ano-genital staining prior to death.

1,750 mg/kg Dose Level (3 animals): All animals died within two days of test substance administration. Prior to death, the animals exhibited piloerection and hypoactivity.

Gross Necropsy Findings:

5,000 mg/kg Dose Level (1 animal): Gross internal necropsy of the decedent revealed the stomach and gastrointestinal tract distended with gas, both red in color. External observations included staining around the anal area.

175 mg/kg Dose Level (2 animals): No gross abnormalities were noted for the two surviving animals when necropsied at the conclusion of the 14-day observation period.

550 mg/kg Dose Level (4 animals): No gross external abnormalities were noted for any of the animals when necropsied at the conclusion of the 14-day observation period. Gross internal necropsy of the animal that died on Day 7 revealed the stomach and gastrointestinal tract distended with gas, both red in color. No other internal gross changes were observed in this animal. No gross internal abnormalities were noted for the other decedent or the two surviving animals when necropsied at the conclusion of the 14-day observation period.

1,750 mg/kg Dose Level (3 animals): Only two of the three decedents revealed gross internal abnormalities including stomach distended with gas or stomach slightly red in color. No gross external abnormalities were noted for any of the euthanized animals when necropsied at the conclusion of the 14-day observation period.

Statistical Analysis:
Test performance and calculation of the LD_{50} was conducted according to the Agency's developed software package (AOT425StatPgm).

DATA REVIEW FOR SKIN SENSITIZATION TESTING (OPPTS 870.2600)
(BUEHLER METHOD)

Product Manager: 33
MRID No.: 477075-05

Reviewer: CSC and Ian Blackwell
Completion Date: January 12, 2009
Project ID: 08-124-5

Testing Laboratory: Tox Monitor Laboratories, Inc., Oak Park, IL
Author: Michael Kukulinski, B.S., L.A.T.G.

Quality Assurance (40 CFR §160.12): A Quality Assurance (QA) statement was included. A statement of Good Laboratory Practice (GLP) compliance was included stating that this study was conducted in accordance with the U.S. EPA Good Laboratory Practice Standards, 40 CFR 160.

Test Material: Maquat 25:12, 25% Glutaraldehyde, 12.5% active quat
Lot #: 1621-224 / Clear liquid

Positive Control Material: 1-Chloro-2,4-dinitrobenzene
Historical data – (date of historical positive control test was not provided)

Species: 32 Guinea pigs; Hartley, albino
Sex: Range-Finding: 2 Males
Test Group: 20 Males
Naïve Control Group: 6 Males
Naïve Control Group – Rechallenge: 4 Males (not required)
Age: Young adult (specific age not reported)
Weight: 404-491 grams; at study start
Source: Kuiper Rabbitry, Gary, IN
Housing: Temperature Range: 63-73°F (17-23°C)
Humidity Range: 30-70%
Photoperiod: 12-hour light/12-hour dark cycle
Acclimation: At least 5 days

Method: Buehler Method

Summary:

1. **Based on these findings and on the evaluation system used, Maquat 25:12, 25% Glutaraldehyde, 12.5% active quat is not considered to be a contact sensitizer.**

2. **Classification:** Supplementary

Procedure (Deviations from 870.2600):

- The laboratory study states that historical positive control data are presented in Appendix 1; however, Appendix 1 was not provided.
- The guidelines recommend using at least 10 animals as controls. The laboratory used 6 animals as controls, reserving 4 animals for re-challenge if necessary.

- The guidelines state that, as a minimum, the erythema and edema must be graded. The laboratory only graded erythema.
- The scoring scale provided in Addendum 1 of the laboratory study appears to be unnecessary, as pages 12 and 16 of the laboratory study present a scoring scale.

Procedure:

Preliminary Irritation Testing: The irritation phase had the purpose of determining the irritation potential of the test material. The irritation potential of the test material at levels of 12%, 6%, 3%, and 1.5% were evaluated in one group of two animals. Four levels of test material were evaluated per animal. Dilutions of the test material were formulated w/w in distilled water. The position of the different concentrations of the test materials on the animals was varied to adjust for possible site-to-site variation in response.

Closed patches were applied to the animals in the following manner: A 0.4 mL quantity of each test preparation was applied directly into a 25 mm Hilltop Chamber. The animals were held gently, and the chambers were applied as quickly as possible to the clipped left shoulder. The chambers were secured with Micropore tape and further secured with Kendall adhesive tape. Approximately six hours later, the tape and chambers were removed. The day following the irritation exposure, all animals were scored according to a scoring system provided in the laboratory report. The scoring was repeated the following day.

Based upon the irritation screen results, the test material was dosed as 3.0% concentration in distilled water for the induction phase of the study and as 1.5% concentration in distilled water, the highest non-irritating concentration, for the challenge phase of the study.

Preparation and Selection of Animals: The day prior to test material exposure, the hair was removed from each of the animal's backs using a small animal clipper. Only animals with skin sites that were free from defects or alterations in coloration or texture were used.

Induction Phase: The purpose of this phase was to dermally expose the animals to the test material so that, if the material is a sensitizer, the physiological process required to ultimately allow the generation of an immunological response can be initiated. The left shoulder of each test animal was clipped with a small animal clipper the day before exposure. The animals were held gently and the chambers were applied as previously described. Two additional induction doses were conducted following the same procedure, at weekly intervals. After the last induction exposure, the animals were left untreated for two weeks (14 days) before primary challenge. The day following each induction exposure, all animals were scored according to the scoring system. The scoring was repeated the following day. For reporting purposes, the first and second gradings were designated as 24- and 48-hour readings, respectively.

DATA REVIEW FOR ACUTE DERMAL TOXICITY TESTING (OPPTS 870.1200)

Product Manager: 33
MRID No.: 477075-04

Reviewer: CSC and Ian Blackwell
Completion Date: December 23, 2008
Project ID: 08-124-4

Testing Laboratory: Tox Monitor Laboratories, Inc., Oak Park, IL
Author: Michael Kukulinski, B.S., L.A.T.G.

Quality Assurance (40 CFR §160.12): A Quality Assurance (QA) statement was included. A statement of Good Laboratory Practice (GLP) compliance was included stating that this study was conducted in accordance with the U.S. EPA Good Laboratory Practice Standards, 40 CFR 160.

Test Material: Maquat 25:12, 25% Glutaraldehyde, 12.5% active quat
Lot #: 1621-224 / Clear liquid

Dosage: 2,000 mg/kg (applied neat)

Species: 10 Rabbits; New Zealand, albino
Sex: 5 Males and 5 Females. Females were nulliparous and non-pregnant.
Age: Young adult (at least 12 weeks old)
Weight: 2.34-3.22 kilograms
Source: Kuiper Rabbitry, Gary, IN
Housing: Temperature Range: 63-73°F (17-23°C)
Humidity Range: 30-70%
Photoperiod: Light-controlled room
Acclimation: At least 5 days

Summary:

1. **Acute Dermal LD_{50} (mg/kg)**: Male and Female Rabbits: >2,000 mg/kg

2. **The estimated acute dermal LD_{50} is** greater than 2,000 mg/kg in male and female rabbits.

3. **Toxicity Category**: III **Classification**: Acceptable

Procedure (Deviations from 870.1200):

- No procedure deviations were reported.
- The guidelines state that body weight changes should be calculated and recorded when survival exceeds one day. Individual body weights of test animals were recorded; however, body weight changes were not reported.
- The guidelines state that, where lighting is artificial, the sequence should be 12 hours light/12 hours dark. The laboratory did not specify the lighting sequence.

Results:

Reported Mortality

Dose Level (mg/kg)	Number Dead / Number Tested		
	Males	Females	Total
2,000	0 / 5	0 / 5	0 / 10

Observations:
All animals survived exposure to the test substance. Nine animals lost weight through Day 7. Seven animals failed to gain weight by Day 14, as compared to initial body weights. One animal gained weight consistently during the study. All animals exhibited erythema, edema, and necrosis by Day 1 of the study. Erythema cleared by Day 8 in all animals, edema cleared by Day 13 in five animals, and necrosis persisted until final sacrifice.

Gross Necropsy Findings:
Gross external necropsy of the euthanized animals revealed necrosis and/or edema at the application site. No gross internal changes were observed for any of the animals when necropsied at the conclusion of the 14-day observation period.

Challenge Phase: The purpose of this phase was to investigate the elicitation of response to the test material. The test animals, which had three previous exposures to the test material at appropriate intervals, were exposed to the test material in the challenge phase, fourteen days after the last induction exposure. The same exposure procedure as previously described was used, except the Hilltop Chambers were applied to a skin site that had not been previously exposed. Each animal received a single chamber of the test material. The day following the primary challenge exposure, all animals were scored according to the scoring system. The scoring was repeated the following day. For reporting purposes, the first and second gradings were designated as 24- and 48-hour readings, respectively.

Historical Positive Control: [The laboratory study states that historical positive control data are presented in Appendix 1. Appendix 1 was not provided.] Based on the study protocol, positive control animals were treated with a 0.3% dilution of 1-chloro-2,4-dinitrobenzene in 80% aqueous ethanol for induction and with a 0.1% dilution of 1-chloro-2,4-dinitrobenzene in acetone for challenge.

Results:

Induction Phase:

Test Animals (3% concentration of the test substance in distilled water): Slight to moderate erythema (0.5-2) was noted for all test sites during the induction phase.

Historical Positive Control Animals (0.3% dilution of 1-chloro-2,4-dinitrobenzene in 80% aqueous ethanol): Appendix 1 was not provided and, therefore, results were not available.

Challenge Phase:

Test Animals (1.5% concentration of the test substance in distilled water): Slight erythema (0.5) was noted for seven of twenty test sites 24 hours after challenge. Slight but confluent erythema (1) was noted for one of twenty test sites 24 hours after challenge. Irritation had cleared from all test sites by 48 hours.

Naïve Control Animals (1.5% concentration of the test substance in distilled water): Slight erythema (0.5) was noted for three of six naïve control sites 24 hours after challenge. Slight but confluent erythema (1) was noted for one of six naïve control sites 24 hours after challenge. Irritation had cleared from all control sites by 48 hours.

Historical Positive Control Animals (0.1% dilution of 1-chloro-2,4-dinitrobenzene in acetone): Appendix 1 was not provided and, therefore, results were not available.

Historical Naïve Control Animals (0.1% dilution of 1-chloro-2,4-dinitrobenzene in acetone): Appendix 1 was not provided and, therefore, results were not available.

Sensitization Response Indices (Erythema)

	Incidence of Positive Response[1]		Severity[2]	
	Hours		Hours	
	24	48	24	48
Test Animals – Challenge	1/ 20	0 / 20	0.23	0
Naïve Control Animals – Challenge	1 / 6	0 / 6	0.42	0

[1]Animals with scores greater than 0.5

[2]Sum of the erythema scores divided by the number of animals evaluated

Test Animal Group Skin Reaction Scores

Treatment Phase	Induction						Challenge	
	1		2		3			
Concentration	3%		3%		3%		1.5%	
Hours[1]	24	48	24	48	24	48	24	48
Animal No. / Sex								
Test Group								
286 / M	1	0	2	1	2	1	0	0
287 /M	1	0	1	0.5	1	0	0	0
288 / M	1	0.5	1	0	1	0	0.5	0
289 / M	2	1	1	0	2	0	0	0
290 / M	1	0	1	0.5	1	0.5	0	0
291 / M	2	1	2	0.5	1	0	0	0
292 / M	1	0	2	1	2	1	0.5	0
293 / M	1	0	1	0	1	0	0	0
294 / M	1	0.5	1	0	1	0	0.5	0
295 / M	1	0	1	0	1	0	1	0
296 / M	1	0	2	0.5	1	0	0.5	0
297 / M	2	0.5	1	0	1	0.5	0	0
298 / M	1	0	1	0	1	0	0	0
299 / M	2	1	1	0.5	2	0.5	0.5	0
300 / M	1	0.5	2	1	1	0	0	0
301 / M	1	0	1	0	1	0	0.5	0
302 / M	1	0	1	0	1	0	0	0
303 / M	1	0	1	0	1	0	0	0
304 / M	1	0.5	2	0.5	2	0.5	0.5	0
305 / M	1	0	1	0	1	0	0	0
Naïve Control Group								
306 / M	--	--	--	--	--	--	0	0
307 / M	--	--	--	--	--	--	0.5	0
308 / M	--	--	--	--	--	--	0.5	0
309 / M	--	--	--	--	--	--	0	0
310 / M	--	--	--	--	--	--	1	0
311 / M	--	--	--	--	--	--	0.5	0

[1]Hours after induction or challenge dose